FINAL REPORT

MARCH 31, 1991

DSS CONTRACT #23440-9-9300/01-SQ

Entitled: The Extension of Bacterial Plugging to Workover Operations

Respectfully Submitted

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EXECUTIVE SUMMARY

The oil industry uses a very wide variety of particulate matter and of polymers in many of their operations, from the drilling and fracture fluids used early in the history of a well, to the work over fluids used during much of its later productive life. Being trained in chemistry and physics, oilfield engineers have tended to use bulk chemicals to solve particular problems and to ignore the potential usefulness of modern biotechnology. Therefore, initially, oilfield biotechnologists can readily make immediate and very significant contributions to the industry by using their powerful new techniques to replace bulk chemicals with much cheaper and much more efficient biotechnical products.

Our initial discovery in this area (in our 1990-1991 DSS Contract #23440-9-9128-101-SQ) is a case in point. Engineers use particulate material (pecan shells, carbonates, etc.) to plug off the very permeable "thief zones" that cause rapid mud loss and a danger of gas blowout when they are encountered during drilling operations. We can now produce a much more rapid, more complete and more permanent plug in a thief zone by simply adding bacteria and nutrients to the drilling mud whenever mud loss is noted or anticipated. In this contract year, we have accelerated this already rapid technique by combining it with the carbonate particle plugging method used by Canadian Fluid Systems in both thief zone plugging and work over operations, and we can now plug a permeable zone in < 30 minutes. We urgently recommend that this combined technology should be protected by a patent.

This year we expanded our originally stated objectives by addressing the problem of fluid loss from drilling mud when drilling "open hole" through very permeable formations. Engineers now add a very expensive ($30/kg) bulk polymer (polyionic cellulose - PAC) to drilling muds in these circumstances to increase their fluid retention. We have discovered that the growth of certain bacteria in drilling mud allows the use of much cheaper < $2/kg cellulose polymers with much better final fluid retention performance. We urgently recommend that either the bacterium in question, or the polymer that it produces, must be protected with a patent so that we can replace PAC as a drilling mud additive in open hole drilling.

RESUME EXECUTIF

L'industrie pétrolière utilise divers types de particules et de polymères dans plusieurs de leurs opérations, depuis le forage et les liquides de fracture lors des étapes préliminaires d'un puit aux travaux concernant les fluides utilisés lors des phases de production d'un puit.Suivant une formation en chimie et en physique, les ingénieurs pétroliers ont une tendance à concentrer leurs efforts sur l'usage de produits chimiques dans le but de résoudre certains problèmes, et ainsi, à ignorer le précieux potentiel de la biotechnologie moderne. C'est pourquoi, d'instance, les biotechnologistes pétroliers peuvent porter une contribution majeure au remplacement de produits chimiques par des produits biotechniques bien meilleur marchés et beaucoup plus efficaces.

Notre découverte initiale dans ce domaine (Contrat DSS 1990-1991 #23440-909128-101-SQ) est un cas typique. Les ingénieurs utilisent des particules (coquilles de noix, carbonates, etc.) afin de boucher les "zones voleuses" trop perméables qui causent de rapides pertes de boues de forage et posent et un risque de fuite de gaz sous pression lors des opérations de forage. Nous pouvons désormais produire un bouchon bien plus rapide, plus complet, et plus permanent dans une zone voleuse grâce à la simple injection de bactéries et de nutrients dans la boue de forage. Lors de cette année sous contrat, nous avons réussi à accélérer cette technique d'avantage en la combinant avec la méthode de bouchage avec des particules de carbonate utilisée par les Systèmes de Fluides Canadiens. Ceci nous permet de boucher une zone perméable en moins de 30 minutes. Notre recommendation est de protéger d'urgence cette technologie combinée par l'établissement d'une patente.

Cette année, nous avons élargi nos objectifs préliminaires en adressant le problème des pertes de fluides des boues de forage, une difficulté souvent rencontrée lors de forages "ouverts" à travers des zones perméables. En ce moment, les ingénieurs ajoutent un polymère très cher (du cellulose polyionique - PAC, à $30/kg) aux boues de forage afin d'améliorer la rétention de fluides. Nous avons découvert que la croissance de certaines bactéries au sein des boues de forage permet d'utiliser des polymères de cellulose bien meilleurs marchés (moins de $2/kg) et bien plus performants. Nous recommendons que la bactérie en question, ou le polymère qu'elle produit, soit protégé par une patente dans le but de remplacer PAC.

BACKGROUND

Oilfield operations have always been directed by engineers with excellent training in chemistry and physics and these people have always turned to chemical and physical solutions to solve their many day-to-day problems. During drilling and workover operations fluid loss is a recurring problem and plugging and viscosity adjustment have always been achieved by the addition of particles (carbonates, pecan shells) and of synthetic polymers (polyacrylics). This is an excellent area for biotechnology and we have already discovered three areas in which bacterial cells and polymers can replace traditional additives at a fraction of the present cost.

Under a previous DSS Contract (#23440-9-9128-101-SQ) we showed that we could replace pecan shells, fibrous materials, and other detritus currently used to plug thief zones encountered during drilling operations with a more sophisticated bacterial plugging technology. These sand and gravel zones cause catastrophic losses of drilling fluid, which can lead to gas well blowouts, and they must be plugged off rapidly and decisively. We developed two bacterial cultures that, when injected with nutrients, are capable of plugging off sand and gravel formations of >8 darcys in <2 hours. This bacterial "plug" actually enters the thief zone, unlike the detritus plugs that only coat its face, and the bacterial plug therefore stays in place during subsequent drilling operations and obviates the need for expensive cement jobs. We formed a Calgary Mud Loss Group to examine patent protection for this discovery and found that the University of Oklahoma has a patent in this same area. The Group concluded (please see Minutes of the June 19, 1990 meeting - appended) that Canadian Fluid Systems should simply apply for a license to use the University of Oklahoma patent for thief zone plugging. Further it was agreed that Microbios Ltd. and Canadian Fluid Systems should carry out research to combine bacterial plugging with their present carbonate plugging technology and should then apply for a "use patent". This work has now been successfully concluded.

During the June 19, 1990 meeting of the Mud Loss Group it was pointed out that it was current oilfield practice to add very expensive ($30/kg) polyionic cellulose (PAC) polymers to drilling fluid in very permeable rock zones to prevent fluid loss into these zones. We perceived that this fluid loss could be much more economically prevented by the production of microbial polymers within drilling mud and the objectives of this present study were expanded to include this new research (please see Milestone #1 Report-July 30, 1990 and Milestone #2 Report-November 15, 1990).

OBJECTIVES

1. To combine bacterial and carbonate particle plugging technologies to produce a very rapid, very tight, plug.

2. To optimize the bacteria-carbonate plugging technology by the selection of fast-growing glycocalyx-producing bacterial strains and by the development of an economical but effective nutrient package.

3. To determine the concentration of bleach (NaOCl) and the exposure times necessary to completely remove the bacterial component of the bacteria-carbonate plug.

PROGRESS TOWARD OBJECTIVES 1-3

Because of our collaboration with Canadian Fluid Systems, we have had access to their excellent laboratory facilities under the very capable direction of Dr. Alan Jones, and we have been able to use their very realistic plugging cells. These plugging cells can be filled with sand of various permeabilities and they can be run at realistic drilling pressures (>400 psi) and temperatures (30°-50°C). We used these more sophisticated cells to repeat our earlier experiments on plugging with bacteria alone (Pseudomonas and Bacillus species) and we again obtained 99% plugging of a >8 darcy sandpack in <2 hours. When either of our bacterial species were combined with Canadian Fluid Systems carbonate particles of various sizes, the resultant plugging of >8 darcy sandpacks was more complete (>99.9%) and more rapid (<30 minutes). Additionally, we were able to use very fine (<0.2μm) carbonate particles without entry of these particles into the sandpack and resultant "formation damage".

Objective #2 was very rapidly realized because we found that all of a very wide variety (>40 isolates) of environmental organisms produced plugging of >8 darcy sandpacks as well or better than the strains of Pseudomonas and Bacillus used in the original experiments. When we replaced our original nutrient (1/2 strength Brain Heart Infusion medium) with filtered 2% sugar beet molasses we still obtained 99.9% plugging in <30 minutes with the six strains tested. These optimization experiments were conducted in our own laboratories because of logistic considerations, but key experiments were repeated in the Canadian Fluid Systems facility.

In Objective #3 we produced combined carbonate-bacterial plugs, treated them with 5.0% NaOCl, and then tested them for permeability and carbohydrate removal. Treatment with 5.0% bleach for 4 hours removed all of the bacteria and exopolymers from the combined plug (as assayed by CFU counts and the phenol-H_2SO_4 method) and the resultant carbonate plug showed no residues of bacterial structures when examined by Scanning Electron Microscopy (SEM).

SUPPLEMENTARY OBJECTIVE

Tc examine the ability of bacterial polymers to prevent fluid loss

from drilling muds into permeable formations.

PROGRESS TOWARD SUPPLEMENTARY OBJECTIVE

Pseudomonas cells were used in these experiments because of foaming problems with the Bacillus culture. The Pseudomonas were grown in a chemically defined sodium citrate medium at half-strength (1/2 SCM). A 2% Pseudomonas inoculum (1.2 X 10^9 cells/ml) was added to 350 ml of mud in place of the polyionic cellulose (PAC) component.

The ability of a polymer system to prevent fluid loss from a drilling mud into a permeable formation is routinely checked by running a standard API test series. Viscometer readings (centipose) at 600 and 300 rpm are used to calculate the plastic viscosity (P.V.) and yield point (Y.P.) values. The gel or carrying capacity of the mud is calculated from viscosity measurements taken at 600 and 3.0 rpm. The ability of polymers to prevent fluid loss is further tested by pouring 350 ml of the mud onto a #50 Whatman cellulose filter in an API fluid loss cell. Immediately after the application of 100 psi, the amount of fluid lost after one minute (spurt fluid loss) and after 30 minutes (total fluid loss) is measured.

The "ideal" API values for a drilling mud containing PAC (0.5 g) are given in the first line of Table 1. The total fluid loss data from a mud amended with a 2% inoculum of Pseudomonas is shown in Figure 1. A significant reduction in total fluid loss was not noted after 48 hours incubation. Therefore, all muds were allowed to incubate after bacterial inoculation for 48 hours at room temperature prior to viscosity and fluid loss testing. The bacterial inoculum alone was not as effective at preventing fluid loss as was the PAC (0.5 g) only (Table 1). Large numbers of bacteria (4.5 X 10^7/ml) were present in the effluent of the inoculated mud as well. In addition, plastic viscosity and yield point values at 48 hours (Figure 2) were below the desired levels (Table 1) indicating that bacteria alone were not capable of providing the desired mud characteristics.

A 2% bacterial inoculum was, however, capable of reducing the amount of PAC required in the drilling mud. The results obtained with 0.2 and 0.25 g of PAC in conjunction with a bacterial inoculum indicate better fluid loss control (Figure 3) than with 0.5 g of PAC alone. The P.V. and Y.P. values (Table 1) are also closer to the desired API values than when a bacterial inoculum only is used.

More significantly, as shown in Table 1, the combinations of a 2% bacterial inoculum with the less expensive carboxylated methyl cornstarch, cross-linked hydroxypropyl cornstarch and amine-derivatized potato starch (Starpak, FL-7 Plus and Stalok 500, respectively) gave fluid loss control that was as good or much better than the very expensive PAC. In addition, when bacteria were combined with commercial polymers in the drilling mud, much smaller numbers of these organisms were present in the effluent

during fluid loss tests.

Because we propose to produce a bacterial product that will be effective in fluid loss prevention in a wide variety of drilling situations we tested the Pseudomonas strain used in these experiments for halotolerance, pH tolerance, and thermotolerance (Table 2).

Table 2

Tolerance of Pseudomonas Strain

		Maximum Cell Production (cells/ml)
1/2 SCM	Control	1.2×10^9
1/2 SCM	+ 15% NaCl	1.2×10^7
1/2 SCM	at pH 3.0	6.1×10^5
1/2 SCM	at pH 11.0	2.4×10^7
1/2 SCM	at 4°C	4.5×10^7
1/2 SCM	at 21°C	1.7×10^9
1/2 SCM	at 37°C	2.6×10^9
1/2 SCM	at 60°C	1.7×10^7

While we stress that many different organisms will produce polymers that are useful in fluid loss control, it is reassuring that this particular strain would be effective in a very wide range of drilling mud conditions.

CONCLUSIONS AND RECOMMENDATIONS

1. The combination of the bacterial plugging technology (University of Okahoma patent) and the carbonate particle plugging technology (Canadian Fluid Systems patent) produces a very effective, very rapid, very easily controlled, and very economical combined technology that should be protected by a use patent and should be used immediately in the field.

2. It is very clear that bacterial polymers can be used in combination with cheaper cellulose polymers to entirely displace polyionic cellulose (PAC) from the worldwide fluid loss control market. The bacterial polymer may be most efficiently produced in situ in the drilling mud, by inoculation, or it may be most efficiently produced in fermenters and used in the field as an inert additive. This discovery, which has immediate commercial significance, should be protected by patent filing immediately.

3. The oil industry uses many chemical polymeric additives (eg. fracturing fluid viscosifiers) that are expensive and difficult to manipulate. A biotechnology initiative should be launched in the oilfield products area to capitalize on the Canadian scientific lead in this area and to transfer a whole set of new technologies to suitable Canadian companies.

Table 1

STANDARD API - TOTAL FLUID LOSS TESTS

Compound Treatment	600 RPM	300 RPM	Gel	Spurt ml	Total Fluid Loss (ml)	P.V.	Y.P.	Viable Cell Count cells/ml Effluent
PAC-no bugs 0.5 g	22-24	13-15	1/4	< 1.0	8.8-9.2	9.0	6.0	-
Bugs 48 hrs.	13	8	1/8	2.0	15.4	5.0	3.0	4.5×10^7
PAC 0.2 g Bugs 2%	25	15	1/9	< 1.0	9.6	10.0	5.0	1.1×10^6
PAC 0.25 g Bugs 2%	26	15.5	1/9	< 1.0	8.2	10.5	5.0	1.0×10^6
Starpak 2.0 g Bugs 2%	25	16	2/9	< 1.0	6.8	9.0	7.0	1.0×10^6
FL-7 Plus 7.0 g Bugs 2%	25	16	2/9	< 1.0	9.2	9.0	7.0	4.0×10^4
Stalok 500 1.1g + Bugs	≈ 25	16	2/9	< 1.0	≈ 10.0	9.0	4.0	1.2×10^4

Figure Legends

Figure 1: Total fluid loss from a mud amended with a 2% inoculum of Pseudomonas

Figure 2: Plastic viscosity and yield point values from a mud amended with a 2% inoculum of Pseudomonas

Figure 3: Total fluid loss from a mud amended with a 2% inoculum of Pseudomonas and varying amounts of P.A.C.

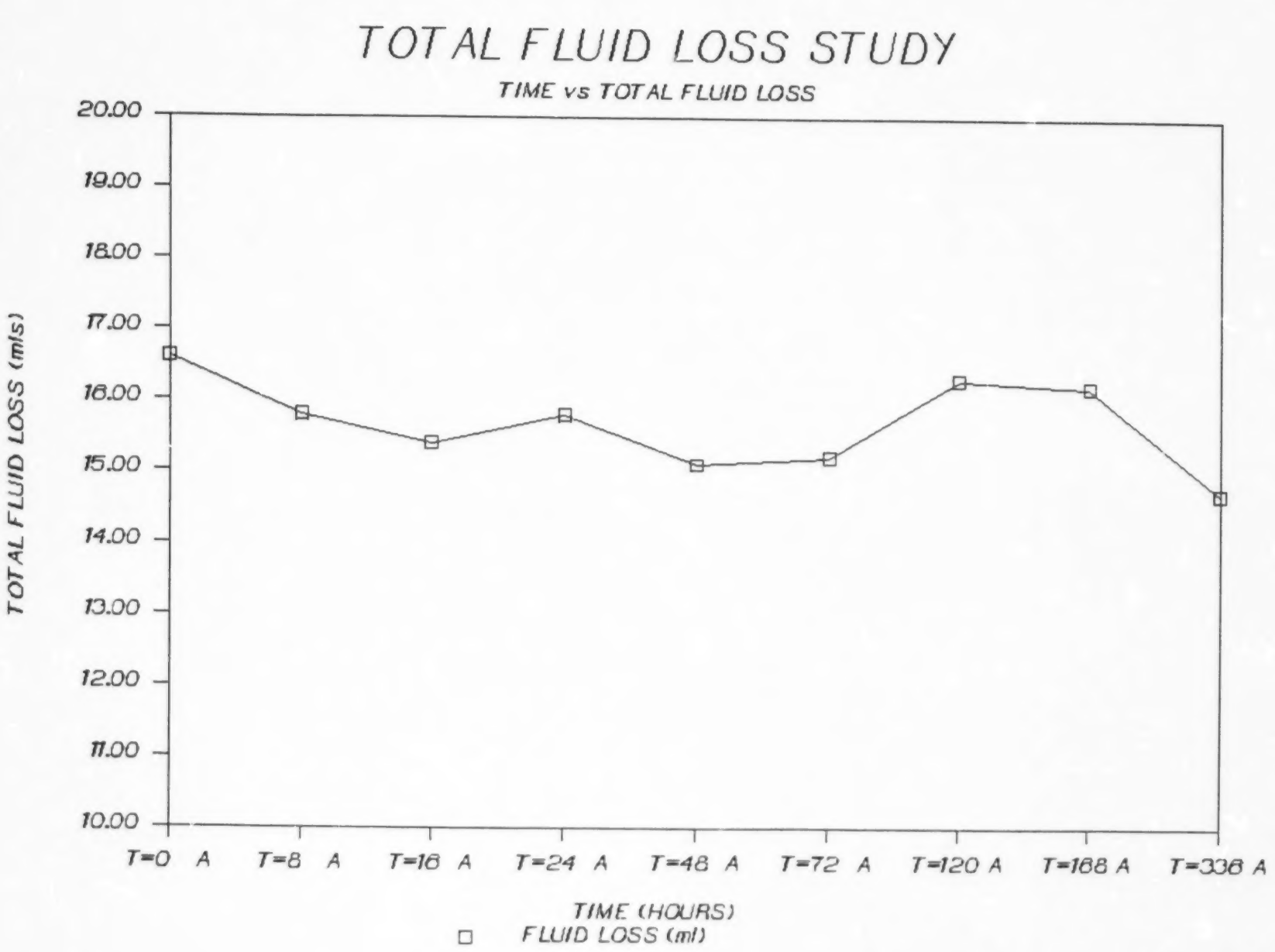
TOTAL FLUID LOSS STUDY
TIME vs TOTAL FLUID LOSS
TOTAL FLUID LOSS (mls)
20.00
19.00
18.00
17.00
16.00
15.00
14.00
13.00
12.00
11.00
10.00
T=0 A
T=8 A
T=16 A
T=24 A
T=48 A
T=72 A
T=120 A
T=168 A
T=336 A
TIME (HOURS)
FLUID LOSS (ml)

Figure 1

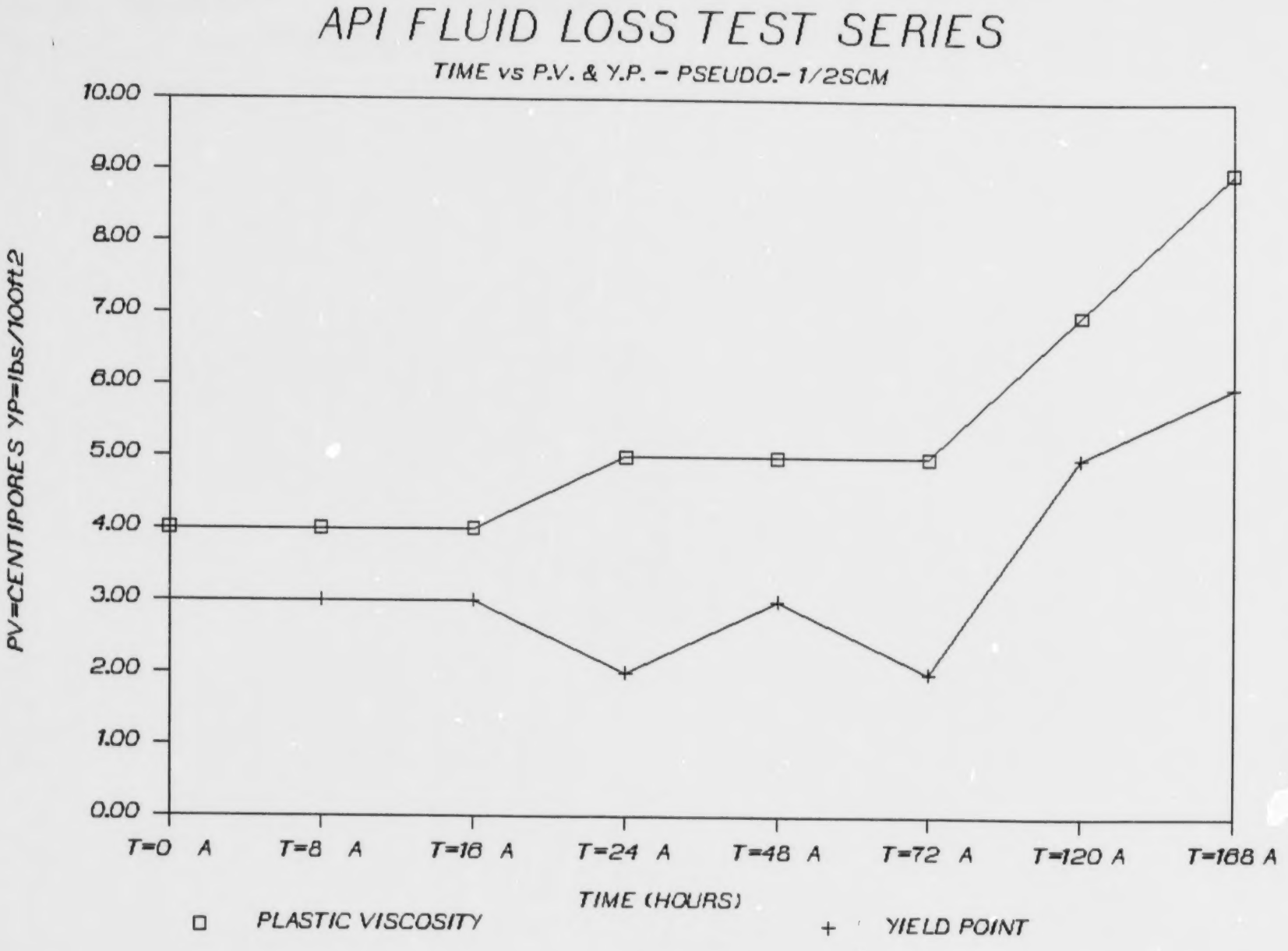
API FLUID LOSS TEST SERIES
TIME vs P.V. & Y.P. - PSEUDO.- 1/2SCM
PV=CENTIPORES YP=lbs/100ft2
10.00
9.00
8.00
7.00
6.00
5.00
4.00
3.00
2.00
1.00
0.00
T=0 A
T=8 A
T=16 A
T=24 A
T=48 A
T=72 A
T=120 A
T=168 A
TIME (HOURS)
PLASTIC VISCOSITY
YIELD POINT

Figure 2

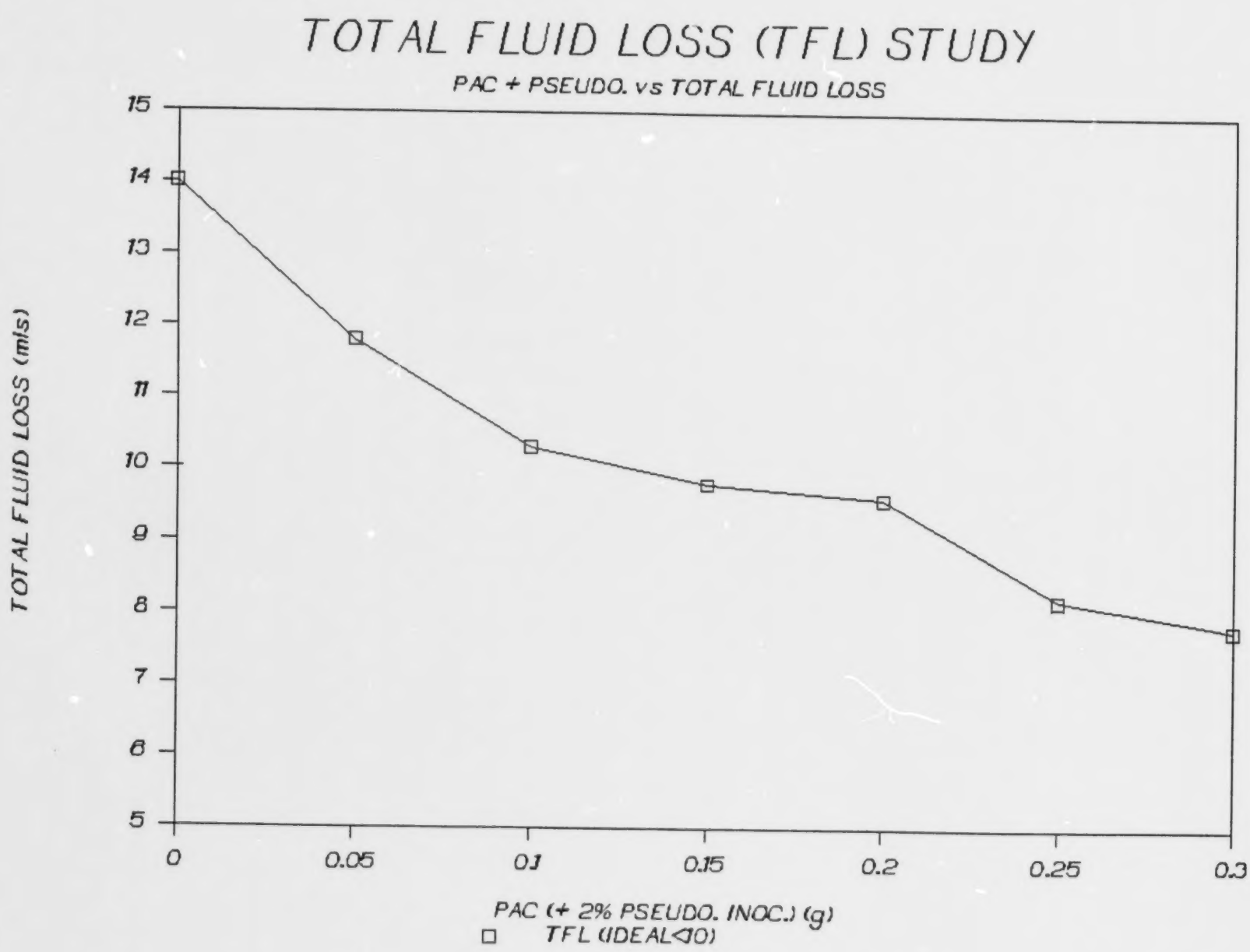
TOTAL FLUID LOSS (TFL) STUDY
PAC + PSEUDO. vs TOTAL FLUID LOSS
TOTAL FLUID LOSS (mls)
15
14
13
12
11
10
9
8
7
6
5
0
0.05
0.1
0.15
0.2
0.25
0.3
PAC (+ 2% PSEUDO. INOC.) (g)
TFL (IDEAL<10)

Figure 3

Appendix

Minutes

Meeting of Calgary Mud Loss Group

June 19, 1990

Present: C. Mark Moennich - B.P. Canada
Larry Pamplin - Canadian Fluids Systems
Bill Costerton - Microbios Ltd. (Chair and Secretary)

All of the parties agreed that, according to their reading of the existing patents in the area of mud loss prevention by bacterial plugging, there does not seem to be an obvious opportunity to file a new patent in this area.

It was moved (C.M.M.) and seconded (B.C.) that "The crown be encouraged again to explore the possibility of filing a patent on some exclusive aspect of the Microbios mud loss plugging technology".

All parties agreed that, in the absence of a new patent covering this technology, no special agreement between the parties and the crown is needed because the obvious course is for Canadian Fluids Systems simply to apply to the University of Oklahoma for a license to use their technology.

All parties agreed that work should begin immediately on the proposed D.S.S. contract with Microbios. This contract will allow Canadian Fluid Ssytems to determine how microbial plugging can best be applied to prevent mud loss in field operations. BP Canada expressed an interest in being kept informed concerning developments in this research, and being acknowledged in publications describing this work, and all parties agreed that this was desirable.

J.W. Costerton

J.W. Costerton
Secretary
Calgary Mud Loss Group

JWC/erm

cc - Dr. R. McCready, CANMET Biotechnology
All attendees